by nolax in a discontinuous procedure by combining polyol and isocyanate. Potential adverse effects of Revcel extracts and degradation products were assessed by according ISO10993-5 measuring total protein content as well as metabolic activity. Cell adhesion was evaluated by cultivating 3T3 mouse fibroblast cells on the material. Revcel is a synthetic, soft, foam-like scaffold with tunable degradation time yet form-stable and that offers optimal pore size for angiogenesis. With the base formulation, extracts showed small adverse effects and cells formed clusters when seeded on foams. Modified formulations affected protein levels to a lesser extent, and exhibited good cell attachment and spreading. Products formed by an *in vivo* like degradation were found to be non-toxic. The results therefore suggest that Revcel has the potential to support wound healing *in vivo*. We acknowledge support by the CTI (project 11874.1 PFLS-LS)

13.P20
Evaluation of the potential of polyhydroxybutyrate-co-hydroxyvalerate bi-layered scaffolds for skin tissue engineering

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Polyhydroxybutyrate-co-hydroxyvalerate (PHB-HV) is natural-based polyester produced by microorganisms under unbalanced growth conditions. This polymer is known for its biocompatibility and biodegradability and has been explored for tissue engineering approaches. The aim of this work was to create a bi-layered construct, based on PHB-HV, for skin tissue replacement. The PHB-HV bi-layered scaffold was produced taking into consideration the two main skin strata, the epidermis and the dermis. For the epidermal fraction, a membrane was developed using a solvent cast approach, while freeze-drying was used to obtain the dermal fraction. The two structures were combined to obtain a scaffold that has a thin compact surface (epidermal) and a porous structure (dermis), as confirmed by scanning electron microscopy (SEM) and micro-CT analysis. Each layer of the scaffold, epidermal and dermal, was respectively seeded with human keratinocytes (Kc) and human dermal fibroblasts (Fb). The characterization by SEM confirmed the typical morphology of Kc and Fb on the expected surfaces. Calcein-AM and DNA quantification showed that both type of cells remained viable along the experiment and were able to attach and proliferate on the respective seeding layers. In conclusion, the developed PHB-HV scaffolds have shown suitable properties to sustain the co-culture of Kc and Fb, which constitutes a solid base for further work to demonstrate its potential for skin replacement.

13.P21
Potential of multiscale fibrous scaffolds for skin tissue engineering

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Mimicking structure, hierarchy and biological functions of native extracellular matrix (ECM) have been one of the major goals of tissue engineering. However, most of scaffold materials for skin tissue engineering highlights on resembling biochemical composition of native tissue. Here we present a rather innovative hierarchical nano/microfibrous chitosan/collagen scaffold, for skin tissue engineering. Scaffolds were developed by sequential electrospinning and freeze-drying. SEM microscopy showed formation of nano/microfibrous layers (~75 nm and ~10 μm fiber diameters). Physico-chemical properties of scaffolds (eg., porosity, tensile strength, swelling behavior, biodegradability) were tested. Scaffolds showed a distinct zone of inhibition against *E.coli*. Scaffolds were evaluated *in vitro* using 3T3 fibroblasts and HaCaT keratinocytes, for assessing matrices' cytocompatibility and cellular response. Presence of type-I collagen in scaffolds encouraged better cell attachment and improved cellular viability. In addition, scaffolds were tested in *ex vivo* human skin equivalent (HSE) model, as a preliminary alternative to *in vivo* animal testing. Results showed migration of keratinocytes along scaffold's surface, causing re-epithelisation of wound-a prerequisite healing and regeneration. Taken together, we observe that by closely mimicking structural/functional attributes of skin, here-in proposed chitosan/collagen scaffolds show great potential for skin tissue engineering application.

13.P22
Healing potential of O2- hydrogels on a full-thickness excision cutaneous wound in diabetic mice

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Foot ulcers and wound healing are problematic for patients with diabetes. At present, many therapies are directed at accelerating wound healing, such as applying electric currents, living skin equivalents, and pharmacological manipulation of factors that modulate the wound healing process. Oxygen (O_2) is a very important mediator associated with wound treatment, and its availability can limit healing rate. We tested the potential effects of O_2-hydrogel therapy on wound healing and regeneration. O_2-hydrogel treatment resulted in accelerated wound closure and formation of granulation tissue in the wound area compared with those in water-hydrogel treated mice. These data suggest that O_2 hydrogels may ultimately provide a novel therapy for accelerating wound healing in patients with diabetes.

13.P23
Comparison of blue and red low level light by LED on healing processes in a skin flap model in rats

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Most studies of low level light therapy have been performed in red or infrared range. We showed the significant impact of blue light to release nitric oxide from nitrosyl complexes. Here we aimed to compare the effects of red and blue light from LED on wound healing in a skin flap model. A skin flap supported by either left or right inferior epigastric neurovascular bundle was illuminated post-OP and on five consecutive days for 10 min with light-emitting diodes (LED) at either 470 nm or 630 nm with 50 mW/cm^2. On day 7 size of necrotic area, flap perfusion, histologic and immunohistochemical parameters were analysed. In both light treated groups wound healing was enhanced, necrotic areas were significantly smaller and flap shrinkage less pronounced compared to controls. Immunohistochemical analyses revealed profound effects of light on neoangiogenesis. In both light treated groups blood vessel count in the perimuscular layer of the skin was twice higher. In the subepidermal layer blue light doubled and red light tripled the amount of blood vessels. Consistently, tissue perfusion was twice higher in both light treated groups as determined by Laser Doppler Imaging. Our data suggest that red and blue light can enhance